(FILE 'HOME' ENTERED AT 09:05:24 ON 03 MAR 2000)

FILE 'MEDLINE, BIOSIS, SCISEARCH, CALLUS' ENTERED AT 09:05:37 ON 03 MAR 2000

L1 351185 (FORMALDEHYDE OR FORMALIN OR GLUTARALDEHYDE OR ALDEHYDE)
L2 1503 L1 AND (NUCLEASE(W) INHIBITOR OR PROTEASE(W) INHIBITOR OR CHELAT ?(W) AGENT)
L3 1786 L1 AND (NUCLEASE(W) INHIBITOR OR PROTEASE(W) INHIBITOR OR CHELAT ?(W) AGENT OR CHELATOR)
L4 1700 L1 AND (NUCLEASE(W) INHIBITOR OR PROTEASE(W) INHIBITOR OR CHELAT ?(W) AGENT OR CHELATOR)

L5 4545 L1 AND (MUREXIDE OR CHROMOTROPIC(W) ACID OR EDTA OR PHENANTHROLI NE OR THIOUREA OR ETHYLENEDIAMINETETRA?)
L6 372 L5 AND (ETHANOL OR BUTANOL OR PENTANOL OR METHANOL OR PROPANOL)
L7 286 DUP REM L6 (86 DUPLICATES REMOVED)

L8 50 L7 AND (RNA OR DNA OR PROTEIN OR TISSUE)
L9 9 L8 AND BUFFER?
L10 17 L7 AND BUFFER?

L10 ANSWER 1 OF 17 MEDLINE
ACCESSION NUMBER: 93065732 MEDLINE
DOCUMENT NUMBER: 93065732
TITLE: Immunohistochemical examination of routinely processed bone marrow biopsies.
AUTHOR: Werner M; Kaloutsi V; Walter K; Buhr T; Bernhards J; Georgii A
CORPORATE SOURCE: Pathologisches Institut, Medizinischen Hochschule Hannover, FRG..
SOURCE: PATHOLOGY, RESEARCH AND PRACTICE, (1992 Aug) 188 (6) 707-13.
Journal code: PBZ. ISSN: 0344-0338.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199302

AB Immunohistochemistry was performed on paraffin sections of 169 bone marrow biopsies fixed in a ***buffered*** ***methanol*** - ***formalin*** solution and decalcified with ***EDTA*** . The biopsies included specimens with normal hematopoiesis, and specimens that were affected by various hematological disorders as well as some metastatic carcinomas. The results demonstrate that a wide spectrum of antigens was preserved in routinely processed bone marrow biopsies, even after long-term fixation up to 12 days. Markers for granulopoietic cells were lysozyme, elastase, DAKO-M 1, and MT 1. Megakaryopoiesis was stained with glycoprotein IIIa, von Willebrand factor, and Ulex europaeus agglutinin (UEA), and erythropoiesis with LN 1. Normal lymphocytes as well as lymphoma cells of all non-Hodgkin's lymphomas tested were positive for leukocyte common antigen (LCA), and at variable degree, for MB 1, 4 KB 5, LN 1, LN 2, UCHL 1, or MT 1. Reed-Sternberg and Hodgkin's cells in Hodgkin's lymphomas were reactive with Ber-H 2, LN 2 and Dako-M 1. In plasma cell disorders, staining for immunoglobulin light chains gave best results. Metastatic carcinomas showed predominantly staining with EMA, and KL 1. A selected

panel of specific cell markers is proposed, which proved to be helpful in routine bone marrow diagnosis in most cases.

L10 ANSWER 2 OF 17 MEDLINE
ACCESSION NUMBER: 86305574 MEDLINE
DOCUMENT NUMBER: 86305574
TITLE: Muconaldehyde formation from 14C-benzene in a hydroxyl radical generating system.
AUTHOR: Latriano L; Zaccaria A; Goldstein B D; Witz G
CONTRACT NUMBER: ES02558
SOURCE: JOURNAL OF FREE RADICALS IN BIOLOGY AND MEDICINE, (1985) 1 (5-6) 363-71.
Journal code: IAJ. ISSN: 0748-5514.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198612
AB It has recently been proposed that muconaldehyde, a six carbon, alpha, beta-unsaturated dialdehyde, may be a nematotoxic metabolite of benzene. The present studies indicate that trans, trans-muconaldehyde is formed from benzene in vitro in a hydroxyl radical (.OH) generating system containing ascorbate, ferrous sulfate and ***EDTA*** in phosphate ***buffer*** , pH 6.7. Muconaldehyde formed from benzene in the .OH generating system was identified by trapping it with thiobarbituric acid (TBA), which results in the formation of an adduct with a 495 nm absorption maximum and a 510 nm fluorescence emission maximum. These maxima were identical to those observed after reacting authentic trans, trans-muconaldehyde with TBA. This finding was supported by thin layer chromatography and solid phase extraction studies. In those studies benzene-derived muconaldehyde cochromatographed with the muconaldehyde/TBA standard. Analyses of the products from the .OH generating system using high performance liquid chromatography (HPLC) confirm that trans, trans-muconaldehyde is a product of benzene ring fission. Regardless of whether or not TBA was used for trapping, samples from the .OH system incubated with benzene contained a peak which cochromatographed with the muconaldehyde standard. The radioactivity profile of fractions collected during HPLC analysis demonstrates 14C-benzene to be the source of the trans, trans-muconaldehyde. The role of hydroxyl radicals in the formation of muconaldehyde was investigated by using dimethyl sulfoxide, mannitol, and ***ethanol*** as .OH scavengers. These scavengers, at concentrations of 10 and 100 mM, were found to cause a dose-dependent decrease in the formation of muconaldehyde. (ABSTRACT TRUNCATED AT 250 WORDS)

L10 ANSWER 3 OF 17 MEDLINE
ACCESSION NUMBER: 86048543 MEDLINE
DOCUMENT NUMBER: 86048543
TITLE: A method for quantitating nanogram amounts of soluble protein using the principle of silver binding.
AUTHOR: Krystal G; Macdonald C; Munt B; Ashwell S
SOURCE: ANALYTICAL BIOCHEMISTRY, (1985 Aug 1) 148 (2) 451-60.
Journal code: 4NK. ISSN: 0003-2697.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals

ENTRY MONTH: 198602

AB A highly sensitive and quantitative assay for measuring protein in solution based on the capacity of protein to bind silver is described. In this procedure, protein samples are first treated with ***glutaraldehyde*** and then exposed to ammoniacal silver. After 10 min, the reaction is terminated by the addition of sodium thiosulfate and the optical density measured at 421 nm. The useful range of the assay for the majority of standard proteins tested lies between 15 and 2000 ng. This represents a 100-fold increase in sensitivity over the Coomassie brilliant blue dye-binding procedure. There is little or no interference from carbohydrates, nonionic detergents, or ***ethanol*** , and pretreatment of protein samples with Bio-Gel P-2 to remove salts, thiol agents, ***EDTA*** , and sodium dodecyl sulfate makes this procedure compatible with most commonly used ***buffers*** . The cost in terms of silver utilization is nominal with a typical assay involving 10 samples tested in triplicate amounting to less than $0.02 U. S.

L10 ANSWER 4 OF 17 MEDLINE
ACCESSION NUMBER: 84018519 MEDLINE
DOCUMENT NUMBER: 84018519
TITLE: Improved procedure for histological identification of osteoid matrix in decalcified bone.
AUTHOR: Yoshiki S; Ueno T; Akita T; Yamanouchi M
SOURCE: STAIN TECHNOLOGY, (1983 Mar) 58 (2) 85-9.
Journal code: V06. ISSN: 0038-9153.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198401

AB Several improvements on the original method of Yoshiki and coworkers for histological identification of osteoid matrix in decalcified bone are described in this report. The first, fixation of bone with neutral ***buffered*** ***formalin*** , a popular and stable fixative, should produce better tissue morphology and ensure easy handling in any laboratory. The second is a simple test for aged cyanuric chloride. Aged reagents show poor or no solubility in ***methanol*** and have almost no effect on differential staining of osteoid matrix. The third is an application of an organic acid solution in place of neutral ***EDTA*** for bone decalcification. Reduced decalcification time with the acid results in rapid preparation of bone sections. Neutral ***formalin*** fixation, immersion in the cyanuric chloride solution, decalcification with an organic acid, and hematoxylin and eosin staining, all quite routine laboratory procedures, yield high quality results for identification of osteoid matrix in bone sections.

L10 ANSWER 5 OF 17 MEDLINE
ACCESSION NUMBER: 82119604 MEDLINE
DOCUMENT NUMBER: 82119604
TITLE: Staining of demineralized cartilage. I. Alcoholic versus aqueous demineralization at neutral and acidic pH.
AUTHOR: Eggert F M; Linder J E; Jubb R W
SOURCE: HISTOCHEMISTRY, (1981 Dec) 73 (3) 385-90.
Journal code: G9K. ISSN: 0301-5564.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198206

AB Demineralization of cartilage with alcoholic ***EDTA*** provides cartilage staining that is no better, as measured by scanning microdensitometry, than that of adequately fixed specimens demineralized with aqueous ***EDTA*** . Aqueous ***EDTA*** is a faster demineralizing agent than alcoholic ***EDTA*** . Certain fixatives can preserve maximal proteoglycan staining in articular cartilage even with subsequent rapid demineralization in formate ***buffer*** at pH 3.3. Although alcoholic ***formalin*** fixation provided optimum quantitative cartilage staining, cetylpyridinium chloride (CPC) in aqueous ***buffered*** ***formalin*** improved cellular detail, but CPC partially suppressed matrix staining.

L10 ANSWER 6 OF 17 MEDLINE
ACCESSION NUMBER: 77140478 MEDLINE
DOCUMENT NUMBER: 77140478
TITLE: Histochemistry of 3beta-hydroxysteroid dehydrogenase in rat ovary. I. Amethodological study.
AUTHOR: Hoyer P E; Anersin H
SOURCE: HISTOCHEMISTRY, (1977 Mar 4) 51 (2-3) 167-93.
Journal code: GPK. ISSN: 0301-5564.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 197707

AB By recording the incubation time needed for initial appearance of the red and blue formazans the reliability of the histochemical method for 3beta-HSD was investigated: 1. Prefixation of small tissue blocks with 1% W/V ***methanol*** -free ***formaldehyde*** (pH=7.2) for up to 30 min preserved morphological integrity as well as maximal enzyme activity. Moreover, the substantivity of formazans and lipids was enhanced. 2. Commercial available ***glutaraldehyde*** (pH=7.2) induced SH groups in the tissue (even at 0.1% W/V for 5 min) thereby enhancing the Nothing dehydrogenase reaction. 3. Preextraction of lipids with acetone for 20 min at -30 degree C caused no loss of activity and was an inevitable step if a reliable activity pattern had to be achieved (e.g. in interstitial cells). 4. No diffusion of enzyme was noticed within 30 min of preincubation in phosphate ***buffer*** (0.2 M, pH=7.2) at 20 degree C. 5. By using the double-section incubation method no diffusion of 3beta-HSD or rediffusion of NADH or PMSH could be noticed withn 45 min of incubation, provided that low concentrations of NAD (0.1 mg/ml) and PMS (0.093 mg/ml) were balanced against the concentration of Nitro BT (0.5 mg/ml) or Tetranitro BT (1.0mg/ml). 6. The utlity of different inhibitors of alkaline phosphomonoesterase was tested and discussed. 7. By inhibiting alkaline phosphomonoesterase with 0.1 mM of L-p-bromotetramisole or 16 mM of beta-glycerophosphate, 3beta-HSD was shown to be exclusively NAD-linked. 8. Levamisole was a potent inhibitor of NADH-tetrazolium reductase as well as 3 beta-HSD, but not of NADPH tetrazolium reductase. 9. 3beta-HSD possess SH groups requisite for the activity as this enzyme was totally inhibited by N-ethyl maleimide. 10. Whether alcohol dehydrogenases may use steroids as substrate is discussed; It is concluded that preextraction (by acetone) and/or the use of an inhibitor of alcohol dehydrogenase (1,10- ***phenanthroline***) has to be performed. 11. Propylene glycol was a poor solvent for all substrates and was itself an excellent substrate for

alcohol dehydrogenase. 12. Specifications for the ideal solvent of steroid substrates in the histochemical practice are proposed. DMSO showed to be promising as a steroid solvent (e.g. extraction of formazans was considerably lower as compared to DMF). 13. The utilization of substrates was descending in the following order (using 1 mM and 0.1 ml/ml of either DMF or DMSO): epiandrosterone, metnandriol, dehydroepiandrosterone and pregnenolone. 14. If DMSO was used as solvent for pregnenolone (but not for the other substrates tested) an evident increase of activity was recorded as compared to DMF.

L10 ANSWER 7 OF 17 BIOSIS COPYRIGHT 2000 BIOSIS
ACCESSION NUMBER: 1996:[illegible]674 BIOSIS
DOCUMENT NUMBER: PREV199699111030
TITLE: Comparative strategies for the purification of a whitefly-transmitted geminivirus infecting tomato fields in Washington and Oregon states.
AUTHOR(S): Abdel-Salam, A. M. (1); Thomas, P. E.
CORPORATE SOURCE: (1) Plant Pathol. Dep., Fac. Agric., Cairo Univ., Giza Egypt
SOURCE: Egyptian Journal of Phytopathology, (1994) Vol. 22, No. 1, pp. 107-123.
ISSN: 0301-8180.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English; Arabic
AB The tomato greenhouse whitefly borne virus (TGWFV) was purified from infected tomato plants. Several described purification techniques for geminiviruses were tested and a recommended technique was reached. The virus was extracted in 0.01 M phosphate ***buffer*** , pH 7.0, containing 10 mM Na2SO3, ImM ethylene diaminetetra acetate (***EDTA***). The virus was clarified with chloroform and ***butanol*** then concentrated with polyethylene glycol and NaCl. The concentrated virus was subjected to one cycle of differential centrifugation. The virus was suspended in the extraction ***buffer*** plus 0.5% triton X-100 (TX-100) and 0.1% polyvinyl pyrolydone (PVP). The virus was further purified by layering it onto 35-65% CsCl gradients. Purified virus had nucleoprotein properties with A max at 260 nm, A min at 240 nm, and A 260/280 ratio of 1.5. Yield of purified virus was 1.37 mg/100 g tissue. The virus possessed a density of 1.30 g/cm-3 in both C-SCl and C-S23O-4 and a nucleic acid percentage of ca. 16% calculated from the ultra-violet spectrophotometry. The obtained virus occurred mostly in dimers 34 times 21 nm. The purified virus was labile to 1% TX-100 (when added to extraction ***buffer***), ***aldehydes*** , and alkaline pH values. Treatment of partially purified virus with Mg++ ions enhanced the solubility of virus preparations and prolonge the survival of virus for two weeks at 4 C. The obtained virus yield from partially purified virus increased in Mg++ treated viru to 4-5 folds comparing to non-treated virus. However, Mg++-treated virions rendered labile in CsCl and Cs2SO4.

L10 ANSWER 8 OF 17 BIOSIS COPYRIGHT 2000 BIOSIS
ACCESSION NUMBER: 1987:414[illegible] BIOSIS
DOCUMENT NUMBER: BA83:20769
TITLE: EXPERIMENTAL STUDIES IN DEMINERALIZATION AND ITS EFFECTS ON CYTOLOGY AND STAINING OF BONE AND MARROW CELLS.
AUTHOR(S): VILLANUEVA A F
CORPORATE SOURCE: BONE MIN. RES. LAB., HENRY FORD HOSP., DETROIT, MI 48202.
SOURCE: J HISTOTECHNOL, (1986) 9 (3), 155-161.

CODEN: JOHIDN. ISSN: 0147-8885.

FILE SEGMENT: BA; OLD

LANGUAGE: English

AB The three types of bone used in this study were rib, iliac crest, and femoral head. 315 bone samples were variously fixed, demineralized, and histologically evaluated after paraffin embedment. Fixation in 10% ***buffered*** ***formalin*** followed by demineralization proved to be the best procedure for preservation of cellular and morphologic detail. If bone samples were to be demineralized in ethylene diaminetetraacetic acid disodium salt (***EDTA***), 70% ***ethanol*** fixation was better than ***formalin*** . ***Formalin*** fixation before ***EDTA*** caused hardness of bone more than alcohol fixation did, making it difficult to determine the end point of demineralization. No reagent was the ideal reagent for removing mineral salts from bone. Many of the procedures were standardized so the entire process could be timed as one block of activity. An empirical formula for demineralizing bone was proposed; and a reservoir of data as a baseline for monitoring quality control in the laboratory was developed; and the latest microtechniques for bone pathology were examined.

L10 ANSWER 9 OF 17 BIOSIS COPYRIGHT 2000 BIOSIS

ACCESSION NUMBER: 1977:238041 BIOSIS

DOCUMENT NUMBER: BA64:60405

TITLE: DOUBLE STRANDED NUCLEIC-ACIDS FOUND IN TISSUE INFECTED WITH THE SATELLITE OF TOBACCO RINGSPOT VIRUS.

AUTHOR(S): SCHNEIDER I R; THOMPSON S M

SOURCE: VIROLOGY, (1977) 78 (2), 453-462.
CODEN: VIRLAX. ISSN: 0042-6822.

FILE SEGMENT: BA; OLD

LANGUAGE: Unavailable

AB A multicomponent population of RNA was purified from [Phaseolus vulgaris] tissue infected with the satellite of tobacco ringspot virus (S TRSV) that is not present in TRSV-infected tissue or in uninfected tissue. Many of the properties are characteristic of ds[double-stranded]RNA, or the so-called replicative form of small RNA viruses; i.e., a sharp melting profile at relatively high temperature and high hyperchromicity and buoyant density in cesium sulfate; the RNA are infective only after denaturation and quick quenching, followed by addition of TRSV. The infectivity is not destroyed prior to denaturation either by pancreatic RNase (in high ionic strength ***buffer***) or by incubation with ***formaldehyde*** . Other properties are not typical of dsRNA: at least 80% of the RNA elute from CF-11 cellulose columns in ***buffer*** (0.1 M NaCl, 0.05 M Tris, 0.001 M ***EDTA*** , pH 6.9)/ ***ethanol*** mixtures that typically elute ss[single-stranded]RNA but not dsRNA. The RNA is composed of many components, some of which are up to 20 times the mass expected from the known mass of the corresponding RNA found in S-TRSV virions. Pancreatic RNase, at relatively high concentrations, converts these higher-molecular-weight dsRNA into dsRNA of lower molecular weight. These lower-molecular-weight double-stranded components retain a high level of infectivity after denaturation (with added TRSV).

L10 ANSWER 10 OF 17 CAPLUS COPYRIGHT 2000 ACS

ACCESSION NUMBER: 1999:753381 CAPLUS

DOCUMENT NUMBER: 132:9594

TITLE: In situ hybridization method for detecting nucleic acid of pathogens in clinical specimens

INVENTOR(S): Shah, Jyotsna S.; Harris, Nick S.
PATENT ASSIGNEE(S): Igenex, Inc., USA
SOURCE: PCT Int. Appl., 27 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFOPMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9960163	A1	19991125	WO 1999-US11046	19990518

W: AU, CA, JP
PW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE

PRIORITY APPLN. INFO.: US 1998-PV88561 19980518

AB The present invention provides a method for detecting a target nucleic acid fragment directly from a specimen obtained from a patient by in situ hybridization. The method is comprised of several steps which are performed in the listed order. A sample of the specimen is deposited onto a slide. The sample is fixed onto the slide with fixative, the fixative comprising either ***methanol*** -acetic acid at a ratio of from 99:1 to 80:20, or ***formalin*** -acetic acid at a ratio of from 99:1 to 80:20. The nucleic acids of the fixed sample are contacted with a probe complex specific for the target nucleic acid fragment, under conditions appropriate for hybridization. Non-hybridized probe complex is rinsed from the sample. The rinsed sample is stained with Evans Blue. The hybridized probe complex is visually detected by microscopy, with the presence of the probe complex being an indication of the presence of the target nucleic acid fragment. The method can be performed with different hybridization ***buffers*** , several of which are disclosed. The method of the present invention is useful for detecting pathogens in a specimen. Specific probe complexes are disclosed which are useful for detecting pathogens of the species Babesia. The method is useful in detecting nucleic acids from a wide variety of specimens, including serum, plasma, sputum, urine, cerebral spinal fluids, tissue, breast milk, and insects such as ticks.

L10 ANSWER 11 OF 17 CAPLUS COPYRIGHT 2000 ACS
ACCESSION NUMBER: 1999:405118 CAPLUS
DOCUMENT NUMBER: 131:41827
TITLE: Universal collection medium
INVENTOR(S): Lorincz, Attila T.; Tang, Yanlin
PATENT ASSIGNEE(S): Digene Corporation, USA
SOURCE: PCT Int. Appl., 40 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFOPMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9931273	A2	19990624	WO 1998-US26342	19981211
WO 9931273	A3	19991007		

W: AU, BR, CA, JP, NO, SG, US, US, US
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL,

PT, SE

AU 9917232	A1	19990705	AU 1999-17232	19981211
PRIORITY APPLN. INFO.:			US 1997-69426	19971212
			US 1998-70436	19980105
			US 1998-82167	19980417
			US 1997-PV69426	19971212
			US 1998-PV70436	19980105
			US 1998-PV82167	19980417
			WO 1998-US26343	19981211

AB This invention provides a novel universal collection medium for cell collection. The medium allows for the first time the ability to perform cytol. and direct mol. anal. on cells preserved in a single sample. This invention also provides novel methods for analyzing cells to assess human conditions.

L10 ANSWER 12 OF 17 CAPLUS COPYRIGHT 2000 ACS
ACCESSION NUMBER: 1996:424967 CAPLUS
DOCUMENT NUMBER: 125:103887
TITLE: ***Methanol*** and ***formaldehyde*** determination by colorimetry using alcohol oxidase
AUTHOR(S): Fujimori, Keiichi; Kitano, Masaru; Takenaka, Norimichi; Bandow, Hiroshi; Maeda, Yasuaki
CORPORATE SOURCE: Fac. Eng., Osaka Prefect. Univ., Sakai, 593, Japan
SOURCE: Bunseki Kagaku (1996), 45(7), 677-682
CODEN: BNSKAK; ISSN: 0525-1931
DOCUMENT TYPE: Journal
LANGUAGE: Japanese

AB MeOH was oxidized to HCHO with alc. oxidase (EC 1.1.3.13, AO), and the HCHO produced was detd. by colorimetry. The sample contg. MeOH and HCHO was bubbled with 100% O for 15 min at flow rate 8.5 L/min. An aliquot of the sample was mixed with 0.1 mL of phosphate ***buffer*** soln. (pH 7.5, 1/15 M) and 0.25 unit AO, and was allowed to stand at 25.degree. for 15 min. HCHO was detd. by colorimetry with AHMT (4-amino-3-hydrazino-5-mercapto-1,2,4-triazole). In this manner, total concns. of HCHO and MeOH could be detd. On the other hand, only HCHO concn. could be detd. without AO. Thus, the MeOH concn. could be calcd. from the difference between these two concns. The detection limit of the present method for MeOH was 2.08 .mu.M, which was 14 times as sensitive as conventional colorimetry.

L10 ANSWER 13 OF 17 CAPLUS COPYRIGHT 2000 ACS
ACCESSION NUMBER: 1993:401872 CAPLUS
DOCUMENT NUMBER: 119:1872
TITLE: Efficient expression of miniprep plasmid DNA after needle microinjection into somatic cells
AUTHOR(S): Thorburn, Andrew M.; Alberts, Arthur S.
CORPORATE SOURCE: Sch. Med., Univ. California, San Diego, La Jolla, CA, 92093-0636, USA
SOURCE: BioTechniques (1993), 14(3), 356,358
CODEN: BTNQDO; ISSN: 0736-6205
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The use of miniprep plasmid DNA in microinjection expts. was studied. As a test plasmid, a .beta.-galactosidase reporter gene expressed from the strong viral cytomegalovirus (CMV) immediate early promoter was chosen. The plasmid was transformed into Escherichia coli by std. methods and a 5-mL culture was grown overnight. Plasmid DNA was isolated by the alk. lysis miniprep method as described (Sambrook, J., et al., 1989). Since it

is crit. in microinjection expts. that no detergents are present in the sample to be injected, the DNA was pptd. first with an equal vol. of isopropanol, then the pellet was suspended in TE ***buffer*** (10 mM Tris-HCl, 1 mM ***EDTA*** , pH 7.4) and ammonium acetate was added to a final concn. of 2.5 M. Three vols. of ***ethanol*** were then added and a second pptn. was carried out. After washing twice with 70- ***ethanol*** , the nucleic acids were dissolved in injection ***buffer*** (50 mM Hepes-NaOH, 40 mM NaCl, pH 7.4). It is not necessary to remove the bacterial RNA, which acts as a carrier in the injection. After estg. the concn. of the plasmid DNA on an ethidium bromide stained agarose gel, the soln. was adjusted so that the injected plasmid was at a final concn. of about 0.2-0.5 .mu.g/.mu.L in the needle. In order to unambiguously identify the injected cells, an inert marker IgG was added to a final concn. of 5 mg/mL. Injected cells can then be identified later by indirect immunofluorescence. Prior to injection, rat embryo fibroblast cells (REF 52) were grown on glass coverslips in Dulbecco's modified Eagle's medium (DMEM) supplemented with 10% fetal bovine serum. In order to identify the area of cells that were injected, a small circle was scored on the coverslip with a diamond-tipped pen. Cells within the scored area were injected directly into the nucleus. Cells outside the scored area were injected into the cytoplasm in order to test whether direct introduction of the DNA into the nucleus was needed for expression. In the authors hands, 100-200 cells can be successfully injected in 10 min with this system. After incubation for one hour to allow for expression of the .beta.-galactosidase gene, the cells were fixed for 5 min in 3.7% ***formaldehyde*** in phosphate- ***buffered*** saline. .beta.-Galactosidase activity was then detected as previously described (Meinkoth, J. L., et al., 1991). In summary, this procedure shows that expression plasmids made by the alk. lysis miniprep method are able to be expressed after direct needle microinjetion into the nucleus of somatic cells. This will allow the rapid screening of plasmids in this type of assay without it being necessary to isolate highly purified DNA using more tedious protocols.

L10 ANSWER 14 OF 17 CAPLUS COPYRIGHT 2000 ACS

ACCESSION NUMBER: 1988:2898 CAPLUS
DOCUMENT NUMBER: 108:2898
TITLE: A silver-binding assay for measuring nanogram amounts of protein in solution
AUTHOR(S): Krystal, Gerald
CORPORATE SOURCE: Terry Fox Lab., British Columbia Cancer Res. Cent., Vancouver, BC, V5Z 1L3, Can.
SOURCE: Anal. Biochem. (1987), 167(1), 86-96
CODEN: ANBCA2; ISSN: 0003-2697
DOCUMENT TYPE: Journal
LANGUAGE: English

AB A highly sensitive assay was developed for measuring protein in soln. based on the capacity of ***glutaraldehyde*** -treated protein to bind silver. This assay has been made more sensitive, with a lower limit of detection of 5 ng, and more reproducible by supplementing protein samples with SDS to reduce protein loss to glassware. Two procedures have been developed. In one, protein samples are supplemented with both SDS and Tween 20 to yield very steep protein dose-response curves, which allow for more precise protein detns., and very stable color formation, permitting absorbance measurements to be made several hours after the assay has been completed. In the second procedure, protein samples are supplemented with SDS alone which results in a less steep dose-response curve and less

stable color formation but makes the assay substantially more tolerant of interfering substances. Thus, proteins in most commonly used ***buffers*** can be assayed directly with the second procedure without the need for ***buffer*** exchange. The procedure of choice, therefore, depends on the type and concn. of interfering substance. Proteins in ***buffers*** totally incompatible with either assay procedure (e.g., those contg. reducing agents) can be easily ***buffer*** exchanged by centrifugation through 0.2% SDS equilibrated, drained Bio-Gel P-2 beads. The clin. utility of this improved assay is demonstrated by the accurate quantitation of protein in 0.5 .mu.L of samples of human cerebral spinal fluid. This assay should therefore prove esp. useful when a limited amt. of protein is available for quantitation.

L10 ANSWER 15 OF 17 CAPLUS COPYRIGHT 2000 ACS
ACCESSION NUMBER: 1983:462544 CAPLUS
DOCUMENT NUMBER: 99:62544
TITLE: Flow injection analysis using immobilized enzyme reagent
AUTHOR(S): Kojima, Tsugio; Hara, Yoshiaki; Morishita, Fujio
CORPORATE SOURCE: Fac. Eng., Kyoto Univ., Kyoto, 606, Japan
SOURCE: Bunseki Kagaku (1983), 32(4), E101-E104
CODEN: BNSKAK; ISSN: 0525-1931
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Alc. dehydrogenase was immobilized on the inner wall of a narrow-bore glass capillary (internal diam. 0.15-0.46 mm; length, 180-400 cm) by using 3-aminopropyltriethoxysilane and ***glutaraldehyde*** , and the resulting glass capillary was used for EtOH detn. by flow-injection anal. By using a carrier soln. consisting of 0.1 mol/dm3 pyrophosphate ***buffer*** (pH 8.5), 2 .times. 10-3 mol/dm3 NAD, 1 .times. 10-3 mol/dm3 ***EDTA*** , and 1 .times. 10-4 mol/dm3 dithiothreitol and a UV detector at 340 nm, EtOH could be detd. with a relative std. deviation of 0.4 3.6% for 1-20 .times. 10-3 mol/dm3 EtOH.

L10 ANSWER 16 OF 17 CAPLUS COPYRIGHT 2000 ACS
ACCESSION NUMBER: 1978:524353 CAPLUS
DOCUMENT NUMBER: 89:124353
TITLE: Laboratory reagent for use in determining ***ethanol*** in liquids
INVENTOR(S): Bucolo, Giovanni
PATENT ASSIGNEE(S): Calbiochem, USA
SOURCE: Swiss, 3 pp.
CODEN: SWXXAS
DOCUMENT TYPE: Patent
LANGUAGE: German
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
CH 598593	A	19780513	CH 1975-13238	19751013

AB Lab. reagents for detn. of EtOH [64-17-5] in body fluids comprise alc. dehydrogenase [9031-72-5] and a mixt. of NAD [53-84-9], a ***buffer*** , an ***aldehyde*** -intercepting (heavy metal complexing) reagent, and an acid to adjust an aq. soln. of the ***buffer*** and ***aldehyde*** intercepting reagent to pH 8.8-9.2. The ***buffer*** and ***aldehyde*** intercepting reagent can be the same compds., such

as 2-amino-2-hydroxymethyl-1,3-propanediol (I) [77-86-1] or 2-amino-2-methyl-1,3-propanediol [115-69-5]. For example, I 842, succinic acid [110-15-6] 104, ***EDTA*** tetrasodium salt [64-02-8] 26 and NAD 17.2 mg, and 215 unit alc. dehydrogenase were dried over P2O5 and mixed. Before use, the compn. was mixed with 13.5 mL H2O. To conduct the test, 0.1 mL blood serum or saliva was dild. with 4.9 mL H2O, and 0.1 mL of the dil. soln. was mixed with 2.6 mL reagent soln., heated 8-10 min at 30.degree., and analyzed by UV spectrometry at 340 nm.

L10 ANSWER 17 OF 17 CAPLUS COPYRIGHT 2000 ACS
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TITLE: Acetaldehyde formation during deproteinization of human blood samples containing ***ethanol***
AUTHOR(S): Stowell, Allan R.; Greenway, Robert M.; Batt, Richard D.
CORPORATE SOURCE: Dep. Chem., Biochem., Biophys., Massey Univ., Palmerston North, N. Z.
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AB A major problem in studying human acetaldehyde (I) metab. is the nonenzymic oxidn. of EtOH to I that occurs during venous blood sample processing for I detn.; therefore, the quant. aspects of the EtOH conversion reaction in venous blood were studied with a new semiautomated method for I detn. in the presence of alc. I was detd. fluorometrically by a modification of the enzymic method of K.E. Crow (1975) in which the original diffusion step was replaced by a vapor-phase transfer using a current of N in a continuous-flow manifold based on that of R. E. Duncombe and W. H. Shaw (1966). Samples (2 mL) were distd. into pyrophosphate ***buffer*** (pH 9.3), and ***buffer*** contg. I was collected into cuvettes from a 2nd gas-liq. separator of the distn. app.

Aldehyde dehydrogenase and NAD were added to each sample and incubated for 15-20 min at room temp. to convert I to HOAc. The NADH formed was directly proportional to .ltoreq.50 .mu.M I. Excitation and emission wavelengths were 350 and 460 nm, resp. Human blood or plasma samples with added EtOH were deproteinized with ice-cold HClO4. Very little or no I was found in aq. EtOH stds. treated with HClO4 or HCl. The extent of I prodn. was variable, related to blood EtOH concn., and could be decreased by increasing the dilu. of blood with the deproteinizing soln. ***Thiourea*** (25 mM) had no effect on I prodn., and insignificant quantities of I were formed during deproteinization of plasma in the presence of EtOH. Apparently, >90% of the I produced during the processing of EtOh-contg.human blood originates from reactions occurring when blood cells, distinct from plasma, are treated with deproteinizer.